CERTIFICATE OF MAILING
I hereby certify that this correspondence is being deposited with the United States Postal Service first class mail in an envelope addressed to: MAIL STOP AF, Commissioner for Patents, P.O. Box 1450, Alexandria, VA 22313-1450,
on October 29, 2010

QUINE INTELLECTUAL PROPERTY LAW GROUP, P.C.

By: Deborah Barragan
Deborah Barragan

OPAP IAPB NOV 0 1 2010 PATENT & TRADEMARK OFFICE

Appl. No.	:	10/536,885	Confirmation No. 6613
Applicant	:	Ebrahim Firoozabady, et al.	
Filed	:	May 31, 2005	
TC/A.U.	:	1638	
Examiner	:	Russell Kallis	
Docket No.	:	63-000600US	
Customer No.	:	22798	

MAIL STOP AF
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION OF DR. EBRAHIM FIROOZABADY PURSUANT TO 37 C.F.R. § 1.132

I, Dr. Ebrahim Firoozabady declare:

I am an expert in the field of plant transformation. My C.V. was attached to my prior declaration filed on April 6, 2009.

I have read the Office Action of June 29, 2010. It is my opinion that the method disclosed in Sripaoraya et al. (Annals of Botany (October 2001) 88: pp. 597-603) ("Sripaoraya") relates to *indirect* regeneration of a plant from transformed leaf base tissue. In other words, subsequent to transformation, the leaf base tissue proceeds through an undifferentiated intermediate prior to shoot production. This is readily observable in the morphological features of shoot primordia shown in Figure 1B of Sripaoraya. I have the original reprint with color photos in my possession. The *undifferentiated* tissues have developed first from the leaf explants. Then, the

undifferentiated tissues have produced two shoot primordial, which are sitting on the top of the undifferentiated tissues and have no apparent connection to the original leaf base explant. It is also apparent that there are no connections of the shoot primordia to the leaf explant through the vascular tissues, which could be confirmed easily by manual separation of the shoot primordia from the undifferentiated tissues. The production of transformed shoots via undifferentiated tissues (indirect regeneration) has been confirmed via personal communications with the lead author, Sripaoraya, whom I have met personally on several occasions at international pineapple conferences. One of skill in the art recognizes that during direct regeneration, most of the tissues of the shoot primordia, including vascular tissues, are directly connected to the explant.

Also, those of skill in the field of plant cell/tissue culture, and specifically pineapple cell/tissue culture, recognize that the combination of components in the culture medium (MSDB medium containing 1 mg/L each of 2,4-D and BAP) used in the method of Sripaoraya will produce an undifferentiated intermediate during the regeneration process (i.e., indirect regeneration).

In contrast to Sripaoraya and as detailed in my prior declaration of April 6, 2009, our method uses *direct* regeneration of transformed shoots from culture of leaf bases. The leaf bases produce shoots within a few weeks without any tissue dedifferentiation.

The pictures attached as Appendix A to the office action response dated April 6, 2009 help illustrate our unique method. As shown in the figures, leaf bases are prepared, inoculated with Agrobacterium and, after cocultivation, are exposed to selection. During the selection, original leaf bases turn brown and die. However, meristems produce transformed shoots directly without any dedifferentiation.

The pictures of Appendix A further illustrate our method: **Fig 1.** Leaf bases after separation from the shoot have meristems attached to them or have meristem regions developed at their bases. **Fig 2 to 4.** The meristems develop into shoots directly. **Fig. 5.** A GUS assay on one of the meristems, which shows transformation.

Further, the transformation of embryogenic callus taught in my U.S. Patent No. 5,952,543 cannot be applied to my direct transformation method described in the current application. The embryogenic callus method transforms individual cells, in

which individual cells produce individual plants. I applied this method to my direct method of regeneration many times, the results of which were production of some sections which died rapidly because chimeric tissues cannot survive the selection to produce transgenic plants. I was well familiar with my embryogenic callus method and the method of Sripaoraya et al., but I could not combine the two methods together to produce a single transformed shoot or shoot bud. This is why I had to invent the new procedure described in our application, which bears no resemblance to the above methods.

I further declare that:

All statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Declarant's signature:

Dr. Ebrahim Firoozabady

10/28/2010
Date